

Product Information

Sigma-Aldrich® CVS10DSYS omniPAGE mini and VS20DSYS omniPAGE maxi dual Vertical Electrophoresis Systems

Catalog Numbers **EP1300 and EP1401**
 Store at Room Temperature
 Technical Bulletin AL-260

TECHNICAL BULLETIN

Product Description

The Sigma-Aldrich® CVS10DSYS omniPAGE mini and VS20DSYS omniPAGE maxi dual vertical gel electrophoresis systems are constructed using the latest injection molding manufacturing techniques. This gives a high quality, low cost product with unsurpassed finish, durability, and strength. In addition, the omniPAGE maxi dual vertical gel electrophoresis unit combines convenient ease-of-use features with high resolution separations. Simple set up using ultra soft silicone seals guarantees trouble free glass plate loading and gel casting.

Guidelines and Restrictions

- Maximum altitude – 2,000 m (6,562 ft)
- Temperature range between 4–65 °C
- Maximum relative humidity 80% for temperatures up to 31 °C, decreasing linearly to 50% relative humidity at 40 °C.
- Not for outdoor usage

This apparatus is rated Pollution Degree 2 in accordance with IEC 664. Pollution Degree 2 states: “Normally only non-conductive pollution occurs. Occasionally, however, a temporary conductivity caused by condensation must be expected”.

Components

Systems include tank, lid, gel running module and electrodes, and accessories (see Table 1).

Table 1.

Accessories included with OmniPage mini and OmniPage maxi dual Vertical Gel Electrophoresis Systems

Product	Glass Plates	Combs	Casting Base	Cooling Pack	Cables
CVS10DSYS omniPAGE mini (Catalog Number EP1300)	Plates: 10 × 10 cm EP1312: Notched, 2 mm thick (Pack of 2) EP1316: Plain with bonded 1 mm spacers (Pack of 2) EP1324: Dummy Plate	EP1352: 1 mm thick, 12 sample (Pack of 2)	EP1304: dual caster EP1305: rubber mat EP1306: Screws × 4	EP1311	EP2002 (Pack of 2)
VS20DSYS omniPAGE maxi (Catalog Number EP1401)	Plates: 20 × 20 cm EP1409: Notched, 2 mm thick (Pack of 2) EP1412: Plain with bonded 1 mm spacers (Pack of 2) EP1417: Dummy Plate	EP1435: 1 mm thick, 24 sample (Pack of 2)	EP1402: dual caster EP1403: rubber mat	EP1408	EP2002 (Pack of 2)

Additional components are available (see Appendix).

Refer to the packing lists as soon as the unit is received to ensure all components have been included. The system should be checked for damage when received. Please contact Sigma-Aldrich if there are any problems or missing items.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Please read the entire technical bulletin before using these units.

Preparation Instructions

Setting up the Gel Tank - Fitting Electrode Cables

- Note the position of the lid on the unit. This shows the correct polarity and the correct orientation of the cables. Black is negative and red is positive.
- Remove the lid from the unit.
Note: If the lid is not removed, fitting the cables may result in loosening the gold plug and damage to the electrode.
- Screw the cables into the tapped holes as fully as possible, so there is no gap between the lid and the leading edge of the cable fitting.
- Refit the lid.

Stock Solutions for SDS-PAGE

- 30% Acrylamide Gel Solution:
 - 30.0 g of Acrylamide
 - 0.8 g of *N,N'*-Methylenebisacrylamide
 - Bring the final volume to 100 mL with distilled water.
- 4× Resolving Gel Buffer (1.5 M Tris-HCl, pH 8.8, with 0.4% SDS):
 - To 110 mL of distilled water add 36.4 g of Tris Base
 - Add 8 mL of 10% SDS solution
 - Adjust pH to 8.8 with 1 N HCl
 - Bring the final volume to 200 mL with distilled water
- 4× Stacking Buffer (0.5 M Tris-HCl, pH 6.8, with 0.4% SDS):
 - To 110 mL of distilled water add 12.12 g of Tris Base
 - Add 8 mL of 10% SDS solution
 - Adjust pH to 6.8 with 1 N HCl
 - Bring to a final volume of 200 mL with distilled water
- 4× Tris-Glycine Running Buffer without SDS:
 - 36.0 g of Tris Base
 - 172.8 g of Glycine
 - Bring volume to 3 L with distilled water

- 1× Tris-Glycine Running Buffer with SDS:
 - 750 mL of 4× Tris-Glycine Running Buffer without SDS
 - 30 mL of 10% SDS solution
 - Bring volume to 3 L with distilled water
- 10% Ammonium Persulfate (APS) Solution:
 - 0.1 g of Ammonium Persulfate
 - Bring volume to 1 mL with distilled water
- N,N,N',N'*-Tetramethylethylenediamine (TEMED), Catalog Number T9281: ready to use as neat liquid
- 4× Sample Buffer:
 - 4 mL of glycerol
 - 2 mL of 2-mercaptoethanol
 - 1.2 g of SDS
 - 5 mL of 4× Stacking Buffer
 - 0.03 g of Bromophenol blue
 - Aliquot into 1.5 mL microcentrifuge tubes. Store at -20°C .

Storage/Stability

The units may be stored at room temperature and operated in the temperature range of 4–65 °C.

Procedure

- Assembly of Vertical Gel Casting System
 - Clean a set of glass plates for each gel, first with distilled water and then with 70% ethanol. One set of glass plates constitutes one notched glass plate and one plain glass plate with bonded spacers. When using a triple glass plate sandwich, two notched glass plates are required, one set of free spacers and a set of plain glass plates with bonded spacers. The plain glass plate is positioned outermost, then a notched glass plate, free spacers, and then a second notched glass plate. Alternatively, accessory notch glass plates with bonded spacers are available. All glass plates, modules, and casting base accessories **must be completely dry during set up. Wet components** are more likely to **misalign and cause leaks**.

2. Assemble the glass plates so that the bottom of the glass plates and the spacers are perfectly aligned. For triple plate sandwiches, the free spacers need to be perfectly aligned, which is best performed using a small spacer or comb to push the spacers apart. Notched glass plates with bonded spacers do not need manual alignment.

Note: The **glass plates** with bonded spacers **have an arrow** in the top of the spacers, which are **slightly longer** than the glass plate **to indicate the top**.

3. The slab gel insert contains pressure bars, which impart even pressure onto the glass plates and allow even screw pressure transfer onto the sealing edge of the glass plate ensuring complete sealing. Ensure the pressure bars are adequately open for the thickness of spacer used. The bar can be opened by loosening the screws or by sliding the clamps. When using a triple glass plate sandwich, the pressure bars will need to be in the completely open position.
4. Position the slab gel insert on a flat surface.
Note: **Do not insert the slab gel insert into the casting base** at this stage.
5. Insert the glass plates into the slab gel insert between the pressure bar and the blue gasket, and fully tighten the pressure bar screws in the order top then bottom. Fully tighten the screw for the omniPage mini vertical unit (EP1300), and the screws sequentially and in an even manner for the omniPage maxi vertical unit (EP1401) in the order middle two, top then bottom, making sure not to wobble the unit. When using the slide clamp mini version, simply slide both gates outwards until fully tightened. When only one gel is being run, the dummy plate must be used in the second position and fully tightened. At this stage, **check that the bottom edges** of the spacers and glass plates **are perfectly aligned**.
6. Position the slab gel insert in the casting base such that the cam pins have handles pointing downwards and are located in the insert holes. The top of the gel running module may need to be pushed down very slightly to locate the cam pins.

7. With the cam pin handles facing directly downwards, turn the cam pins fully through 180° or until the insert has tightened onto the silicone mat. It is best to **turn the cams in opposite directions** to each other. **Do not overturn** as this will cause the glass plates to push upwards and **the assembly will be more likely to leak**.

The unit is now ready for gel preparation and pouring.

Notes: Always reverse the silicone mat after casting to avoid indentations from persisting. Never leave the casting up-stand with glass plates tightened into the casting base for long periods of time, as this will also cause indentations in the silicone mat.

The slide clamp version omniPAGE mini vertical unit (EP1300) also includes 4 screws (EP1306). This system can be used either with the slide clamps or screws as preferred by the user. For those that prefer to use the screws rather than clamps, the screws can be simply inserted into the screw holes. The clamps can be removed by placing each clamp in the fully open position and gently bending the clamp upwards from the slanted end. The holding pin will then slowly release and the clamp can be removed.

B. Gel Preparation

It is always advisable to work using stock solutions, which should be prepared beforehand, see Preparation Instructions, Stock Solutions for SDS-PAGE. Stock solutions are more convenient and save time when it comes to gel pouring. For native gel formulations and running conditions, please consult a laboratory manual. The procedure uses the standard stock solutions described in the Preparation Instructions. This procedure should be adjusted if using different stock solutions or gel formulas.

Table 2 shows the total volume of gel solution required. In subsequent tables, amounts of gel and solutions are given for two 1 mm thick gels, so adjustments will subsequently be needed when running single or more than two gels, and for 0.75, 1.5, or 2 mm thick spacers.

E. Gel Pouring - Gels with **Stacking Layers**

1. Insert the comb into the glass plates and mark a point on the glass plates 1 cm below where the comb teeth end. This indicates the level to which to add the resolving gel.
2. Add 15 μL of TEMED to the resolving gel solution for omniPage mini sized gels and 70 μL for omniPage maxi sized gels, and mix well while avoiding the generation of air bubbles.
3. Fill the glass plates, again avoid the generation of any air bubbles. Filling must be performed quickly before the TEMED causes the gel to become too viscous.
4. Overlay the top of the gel carefully with 1 mL of isobutanol, isopropanol, or distilled water. When using distilled water extra care must be taken to ensure there is no mixing with the gel solution.
5. Let the resolving gel polymerize. Usually this takes ~15 minutes but this can vary due to the freshness of the reagents used. If polymerization takes considerably longer than this, use fresher stock solutions, or add more Ammonium Persulfate Solution and TEMED.
6. Prepare the stacking gel solution (see Table 6) from the standard stock solutions, see Preparation Instructions, Stock Solutions for SDS-PAGE.

Table 6.

Preparation of Stacking Gel Solution

Stock Solution	omniPage mini (EP1300)	omniPage maxi (EP1401)
Distilled Water	4.2 mL	16.8 mL
30% Acrylamide Gel Solution	0.65 mL	2.6 mL
4 \times Stacking Gel Buffer	1.6 mL	6.4 mL
10% Ammonium Persulfate Solution	67 μL	176 μL

7. Carefully mix the stacking gel solution; avoid generation of air bubbles.
8. Pour off the overlay liquid and rinse the top of the resolving gel with distilled water.

9. Add 6.7 μL of TEMED to the stacking gel solution for omniPage mini gels and for omniPage maxi gels add 26.8 μL , and mix well using a Pasteur pipette to fill the glass plates up to the top with stacking gel solution.
10. Carefully insert the comb making sure no air bubbles get trapped under the ends of the comb teeth, as these will inhibit sample progression.
11. Allow the stacking gel to polymerize for 30 minutes

F. Gel Pouring - Gels **without Stacking Layers**

1. Add 15 μL of TEMED to the resolving gel solution for omniPage mini sized gels and 70 μL for omniPage maxi gels, and mix well while trying to avoid generating air bubbles.
2. Fill the glass plates, again avoid generation of any air bubbles. Filling must be performed quickly before the TEMED causes the gel to become too viscous.
3. Carefully insert the comb making sure no air bubbles get trapped under the ends of the comb teeth, as these will inhibit sample progression.
4. Let the resolving gel polymerize. Usually this takes ~15 minutes but this can vary due to the freshness of the reagents used. If polymerization takes considerably longer than this, use fresher stock solutions, or add more Ammonium Persulfate Solution and TEMED.

G. Preparation of Denatured Protein Samples

This procedure is for denatured samples. For native samples, please consult a laboratory handbook.

1. Prepare the protein samples for loading. The volume of sample depends on the capacity of the wells (see Gel Combs, Tables 9 and 10).
2. Using a 0.5 mL microcentrifuge tube or other convenient receptacle, combine the protein sample and 4 \times Sample Buffer. It is always advisable to use protein markers in one of the end lanes. These should be prepared according to the manufacturers instructions.
3. Heat the samples in a water bath or heating block for 2 minutes to denature the samples.
4. Centrifuge the samples in a microcentrifuge for 20 seconds at 12,000 rpm. The protein samples are now ready to load.

H. Loading the Samples

1. If desired, fit the cooling pack(s) into the end of the tank. These should be pre-frozen and fitted with the longest side positioned sideways with the end(s) of the tank and pressed into the recess. Alternatively, these can be fitted down the front of the tank. **Never fit these underneath the module in the bottom of the tank** as this will prevent the flow of current through the gel, and cause slow runs and over-heating.
Note: One pack is supplied as standard for both the omniPage mini and omniPage maxi gel units. Additional packs may be purchased if required.
2. Transfer the gel running module containing cast gels into the main tank in the correct orientation as indicated: +ve (positive electrode, red) on the module aligned with +ve (positive electrode, red) on the tank; -ve (negative electrode, black) on the module aligned with -ve (negative electrode, black) on the tank.
3. Fill the outer tank with 1× Running Buffer. Table 7 shows the recommended volumes of 1× Running Buffer required per gel.
4. Load the samples into the wells using a pipette tip taking care not to damage the wells or introduce any air bubbles.
5. Fill any unused wells with 1× Sample Buffer prepared by 4-fold dilution of 4× Sample Buffer with water.
6. It is a good idea to note the orientation and order the samples were loaded. This can be done by noting which samples were loaded adjacent to each electrode.

Table 7.

1× Running Buffer Volumes

1× Running Buffer Volumes	omniPage mini (EP1300)	omniPAGE maxi (EP1401)
Minimum Volumes - Cooling potential is at a minimum, which may affect resolution. Outer Tank is filled to just flood the bottom of the glass plates → Inner tank is filled to above the wells →	250 mL 500 mL	1.2 L 1.8 L
Maximum Volumes - Cooling is high, offering good resolution of samples. Outer Tank is filled to the maximum fill line → Inner tank is filled to above the wells →	1,200 mL 2.8 L	5.6 L 8.4 L
Volumes when using the cooling packs - Cooling is at a maximum. Cooling packs are inserted behind the gels. Outer Tank is filled to the maximum fill line → Inner tank is filled to above the wells →	1,000 mL 2.3 L	4.6 L 6.9 L

I. Running the Gels

1. Fit the lid and connect to a power supply.
2. Consult Table 8 for recommended power supply voltage settings.
3. Turn the power supply off when the loading dye reaches the bottom of the gel; sooner if the proteins are below 4 kDa in size.
4. Remove the gel running module, first emptying the inner buffer into the main tank. Buffer can be re-used but this may affect run quality if continued.
5. Unscrew the glass plates and gently pry apart the glass plates. The gel will usually stick to one of the plates and can be removed by first soaking in buffer and then gently lifting with a spatula.
6. The gel is now ready to be stained with Coomassie® Blue or silver stain, or the proteins in the gel can be transferred to a membrane by electroblotting for specific band identification and further analysis.

Table 8.
Voltage Settings

Recommended Voltages and Resultant Current for 1 mm thick, 12% gels.	omniPage mini (EP1300)	omniPage mini (EP1401)
1 gel	90–225 V 20–45 mA	120–250 V 20–45 mA
2 gels	90–225 V 40–90 mA	120–250 V 40–90 mA
3 gels	90–225 V 60–135 mA	120–250 V 60–135 mA
4 gels	90–225 V 80–180 mA	120–250 V 80–180 mA

References

1. Sambrook, Fritsch, and Maniatis, *Molecular Cloning A Laboratory Manual*, Second Edition, Cold Spring Harbor Laboratory Press, (Cold Spring Harbor, NY: 1989).
2. *Current Protocols in Molecular Biology*, Greene Publishing Associates and Wiley-Interscience, 1989.

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Appendix

Table 9.
Gel Combs - ominiPage mini gel electrophoresis units (EP1300)

Catalog Number	Description	Sample Volume for a 5 mm deep well gel (µL)
EP1339	Comb 1 Prep, 1 Marker, 0.75 mm thick	500
EP1340	Comb 5 sample, 0.75 mm thick	70
EP1341	Comb 8 sample MC , 0.75 mm thick	40
EP1342	Comb 9 sample, 0.75 mm thick	35
EP1343	Comb 10 sample, 0.75 mm thick	30
EP1344	Comb 12 sample, 0.75 mm thick	25
EP1345	Comb 16 sample MC , 0.75 mm thick	20
EP1346	Comb 20 sample, 0.75 mm thick	15
EP1347	Comb 1 Prep, 1 Marker, 1 mm thick	650
EP1348	Comb 5 sample, 1 mm thick	100
EP1349	Comb 8 sample MC , 1 mm thick	60
EP1350	Comb 9 sample, 1 mm thick	50
EP1351	Comb 10 sample, 1 mm thick	40
EP1352	Comb 12 sample, 1 mm thick	35
EP1353	Comb 16 sample MC , 1 mm thick	25
EP1354	Comb 20 sample, 1 mm thick	20
EP1356	Comb 1 Prep, 1 Marker, 1.5 mm thick	1,000
EP1357	Comb 5 sample, 1.5 mm thick	140
EP1358	Comb 8 sample MC , 1.5 mm thick	80
EP1359	Comb 9 sample, 1.5 mm thick	70
EP1360	Comb 10 sample, 1.5 mm thick	30
EP1361	Comb 12 sample, 1.5 mm thick	50
EP1362	Comb 16 sample MC , 1.5 mm thick	40
EP1363	Comb 20 sample, 1.5 mm thick	30
EP1364	Comb 1 Prep, 1 Marker, 2 mm thick	1300
EP1365	Comb 5 sample, 2 mm thick	200
EP1366	Comb 8 sample MC , 2 mm thick	120
EP1367	Comb 9 sample, 2 mm thick	100
EP1368	Comb 10 sample, 2 mm thick	80
EP1369	Comb 12 sample, 2 mm thick	70
EP1370	Comb 16 sample MC , 2 mm thick	50
EP1371	Comb 20 sample, 2 mm thick	40

MC denotes Multi-Channel pipette compatible.

Table 10.
Gel Combs - ominPage maxi gel electrophoresis units (EP1401)

Catalog Number	Description	Sample Volume for a 5 mm deep well gel (µL)
EP1423	Comb 1 Prep, 1 Marker, 0.75 mm thick	1,100
EP1424	Comb 5 sample, 0.75 mm thick	160
EP1425	Comb 10 sample, 0.75 mm thick	80
EP1426	Comb 18 sample MC , 0.75 mm thick	40
EP1427	Comb 24 sample, 0.75 mm thick	30
EP1428	Comb 30 sample, 0.75 mm thick	25
EP1429	Comb 36 sample MC , 0.75 mm thick	20
EP1430	Comb 48 sample, 0.75 mm thick	15
EP1431	Comb 1 Prep, 1 Marker, 1 mm thick	1,500
EP1432	Comb 5 sample, 1 mm thick	200
EP1433	Comb 10 sample, 1 mm thick	100
EP1434	Comb 18 sample, 1 mm thick	50
EP1435	Comb 24 sample, 1 mm thick	40
EP1436	Comb 30 sample, 1 mm thick	35
EP1437	Comb 36 sample MC , 1 mm thick	25
EP1438	Comb 48 sample, 1 mm thick	20
EP1439	Comb 1 Prep, 1 Marker, 1.5 mm thick	2,200
EP1440	Comb 5 sample, 1.5 mm thick	320
EP1441	Comb 10 sample, 1.5 mm thick	160
EP1442	Comb 18 sample, 1.5 mm thick	80
EP1443	Comb 24 sample, 1.5 mm thick	60
EP1444	Comb 30 sample, 1.5 mm thick	50
EP1445	Comb 36 sample MC , 1.5 mm thick	40
EP1446	Comb 48 sample, 1.5 mm thick	30
EP1447	Comb 1 Prep, 1 Marker, 2 mm thick	3,000
EP1448	Comb 5 sample, 2 mm thick	400
EP1449	Comb 10 sample, 2 mm thick	200
EP1450	Comb 18 sample, 2 mm thick	100
EP1451	Comb 24 sample, 2 mm thick	80
EP1452	Comb 30 sample, 2 mm thick	70
EP1453	Comb 36 sample MC , 2 mm thick	50
EP1454	Comb 48 sample, 2 mm thick	40

MC denotes Multi-Channel pipette compatible.

Table 11.
Additional Equipment

Product Description	Catalog Number
Parafilm® M roll size 2 in. × 250 ft	P7543
Parafilm M roll size 4 in. × 125 ft	P7793
Parafilm M roll size 4 in. × 250 ft	P7668
Screws × 4	EP1306
Cables (Pack of 2)	EP2002
Cooling Packs	EP1311
Gel Staining Tray	T0567
Transilluminator UV/White light AC input 115 V, 60 Hz	Z363820
Power supply 250V AC input 110 V US 3-pin plug	PS2501
Power supply 250V AC input 220 V EU 2-pin plug	PS2502

Table 12.
Electrophoresis Chemicals

Product Description	Catalog Number
Sodium Dodecyl Sulfate (SDS), for electrophoresis, ≥98.5% (GC)	L3771
Tris-Glycine-SDS Buffer, 10× Concentrate	T7777
Trizma® Base, primary standard and buffer, >99.9% (titration)	T1503
Glycine, for electrophoresis ≥99%	G8898
Ethanol, 200 proof (absolute), for molecular biology	E7023
2-propanol, BioReagent, for molecular biology ≥99%	I9516
Hydrochloric acid solution, 1.0 N	H3162
Acrylamide, for electrophoresis ≥99% (HPLC), powder	A3553
Acrylamide solution, 40% for electrophoresis, sterile-filtered	A4058
<i>N,N'</i> -Methylenebisacrylamide, for electrophoresis ≥98%, powder	M7279
<i>N,N'</i> -Methylenebisacrylamide solution, for electrophoresis, 2.0% in water	M1533
Ammonium Persulphate, for electrophoresis ≥98.0%	A3678
Water, Molecular Biology Reagent	W4502
2-Mercaptoethanol, for electrophoresis	M7154
TEMED, <i>N,N,N',N'</i> -Tetramethylethylenediamine, BioReagent, suitable for electrophoresis, ~99%	T9281
Glycerol ≥99%, for electrophoresis	G8773
Bromophenol Blue, sodium salt, for electrophoresis	B5525
EZBlue™ Gel Staining Reagent	G1041
Brilliant Blue R Staining Solution, ethanol solution	B6529
ProteoSilver™ Plus Silver Stain Kit	PROTSIL2
ProteoSilver Silver Stain Kit	PROTSIL1
Gel Loading Solution, Type I, for non-denaturing PAGE	G7654

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